Physicochemical and Microbiological Stability of Compounded Clonidine Hydrochloride Oral Liquid Dosage Forms in PCCA Base, SuspendIt™

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INTRODUCTION AND AIMS

Clonidine Hydrochloride is a centrally acting alpha-agonist hypotensive agent available as tablets for oral administration in three dosage strengths: 0.1 mg, 0.2 mg and 0.3 mg.

No commercial liquid dosage form of clonidine hydrochloride currently exists. An extemporaneously compounded suspension from pure drug powder or commercial tablets/capsules would provide an alternative option to meet unique patient needs. The purpose of this study was to determine the physicochemical and microbiological stability of extemporaneously compounded clonidine hydrochloride suspensions in the PCCA base SuspendIt™. This base is a sugar-free, paraben-free, dye-free, and gluten-free thixotropic vehicle containing a natural sweetener obtained from the monk fruit. It thickens upon standing to minimize settling of any insoluble drug particles and becomes fluid upon shaking to allow convenient pouring during administration to the patient. The study design included two concentrations to provide stability documentation over a bracketed range for eventual use by compounding pharmacists.

A robust stability-indicating HPLC assay for the determination of the chemical stability of clonidine hydrochloride in SuspendIt™ was developed and validated. Suspensions of clonidine hydrochloride were prepared in PCCA SuspendIt at 20-mcg/mL and 100-mcg/mL concentrations, selected to represent a range within which the drug is commonly dosed. Given the potent nature of the drug, a 2% triturate of clonidine hydrochloride in microcrystalline cellulose was used to prepare the samples. Samples were stored in plastic amber prescription bottles at two temperature conditions (5^0C and 25^0C) and assayed over an extended period of time. Physical data such as pH, viscosity, and appearance were also noted. Samples were also tested for microbiological stability. The goal was to provide a viable, compounded alternative for clonidine hydrochloride in a thixotropic liquid dosage form, with an extended beyond-use-date to meet patient needs.

METHODS

Development of a stability-indicating HPLC assay method for Clonidine Hydrochloride

The HPLC analytical method developed was demonstrated to be stability indicating by subjecting clonidine hydrochloride samples to accelerated degradation. A forced degradation study was performed to determine if any degradants interfered with the analytical peak for clonidine hydrochloride. These forced degradations included caustic, acidic, peroxide, and ultraviolet light degradation. For the caustic degradation, 0.5 mL of 5N NaOH was added to a 5-mL volumetric flask containing either 50 µL of 100-µg/mL stock solution of clonidine hydrochloride in mobile phase, or approximately 50 mg of a test formulation containing 100 µg/mL clonidine hydrochloride in PCCA SuspendIt. The sample was heated to 60°C for 1 hour. For the acidic degradation, the stock solution or formulation was mixed with 0.5 mL of a 1N HCl solution and stored at room temperature for 1 hour. The peroxide degradation was accomplished in an analogous manner by mixing the sample or stock solution with 483.3 µL of deionized water and 16.7 µL of a 30% hydrogen peroxide solution, resulting in a 1% peroxide solution. Forced degradation by UV light was achieved by placing either 100-µg/mL stock solution, or 50 mg of the 100-µg/mL formulation in 5-mL volumetric flasks in a Millipore UV sterilizer (Catalog No. XX6370000, Billerica, Massachusetts) for 1 hour.

Preparation of Clonidine Hydrochloride Suspensions in SuspendIt™

Two suspensions, one containing 20-µg/mL, and the other containing 100-µg/mL of clonidine hydrochloride in PCCA SuspendIt were prepared by first weighing out either 1.0 gram or 5.0 grams respectively, of a 2% triturate of clonidine hydrochloride in microcrystalline cellulose and placing the powder in a mortar. The powder was levigated to a smooth paste using a small amount of PCCA SuspendIt. Additional PCCA SuspendIt was added to the mortar and the contents transferred into a 1000-mL volumetric flask using a rubber spatula.

For both drug concentrations, twelve 4-oz. amber plastic prescription bottles were filled with 80 mL of the prepared suspension, retaining the remainder for initial (zero day) analysis. The specific gravity was determined to be 1.0 for both the 20-µg/mL, and the 100-µg/mL batches. Six bottles containing 80 mL of suspension each were sealed, wrapped in parafilm, divided into two groups of three bottles each, and stored either at room temperature (25°C) in a desiccator, or under refrigerated conditions (5°C). The temperature at each storage location was monitored throughout the study. Samples from each temperature and concentration were analyzed and characterized initially on day zero, and subsequently after 7, 14, 28, 42, 63, 91, 119 and 182 days of storage. The remaining six bottles containing 80 mL each of the suspension were tested for microbiological stability.

Analysis and Characterization of Clonidine Hydrochloride Suspensions

Samples of clonidine hydrochloride in SuspendIt™ were analyzed on a Waters chromatographic system (Waters Corporation, Milford, Massachusetts) using a 717 autosampler, a 600-quaternary pump, and a 996-photodiode array detector set at 220 nm for the detection of the clonidine hydrochloride. An isocratic mobile phase containing 95% v/v of a 50 mM sodium phosphate dibasic solution adjusted to pH 7.9 using phosphoric acid and 5% v/v acetonitrile was used at a flow rate of 1.0 mL/min. The injection volume was 30.0 microliters. An Xbridge C18 2.1 X 100-mm 5-µm particle size column was used for the separation.

A series of standards ranging from 0.25 µg/mL to 2.0 µg/mL were prepared in mobile phase from a 10.0 µg /mL stock solution of clonidine in mobile phase prepared fresh for each sampling period. The stock solution was prepared from a 2% triturate of clonidine hydrochloride in microcrystalline cellulose by weighing out 25.00 mg of the triturate and dissolving the drug in mobile phase in a 5-mL volumetric flask. After the drug was dissolved, the solution was centrifuged to remove the cellulose and then diluted 1:10 in mobile phase, producing a stock solution of 10-µg/mL This concentration range was chosen to bracket the target concentration (1 µg/mL) of the samples. Chromatograms were acquired for the standards and samples. A least squares analysis was performed on the calibration curve using the peak areas from the clonidine hydrochloride peaks at 220 nm to determine the clonidine concentrations of the samples. Using the initial specific gravity measurements of the formulations, the weight/weight measurements were converted to weight/volume units.

The suspensions were also analyzed for pH, appearance and intrinsic viscosity. The pH of each sample was measured on a VWR Scientific pH meter using an Ag/AgCl combination electrode, calibrated prior to analysis. The viscosity was determined using a Brookfield DV-III Ultra programmable cone/plate rheometer fitted with a cpe-40 spindle.

RESULTS

The HPLC method utilized in the study clearly separated any peaks associated with the SuspendIt™ from the analytical peak for the clonidine hydrochloride (Figure 1). The method also displayed good linearity over the observed concentration range (Figure 2). Forced degradation studies revealed that peaks associated with the degradants had much shorter retention times than the clonidine hydrochloride peak and showed no interference with its analytical peak (Figure 3).

Clonidine hydrochloride formed a translucent suspension in PCCA SuspendIt at both concentrations, with the 100-µg/mL concentration showing a slightly higher opacity. The pH of the 20-µg/mL samples displayed no significant changes over the test period (Table 1). The 100-µg/mL samples, however displayed a slight decrease in pH ranging from 5.12±0.01 at the start of the study to 4.74±0.31 and 4.57±0.09 for the refrigerated and room temperature samples, respectively. The viscosities for both the refrigerated and room temperature samples at both concentrations stayed fairly consistent throughout the test displaying no signs of breakdown (Table 2). The viscosity was sufficient to avoid caking and obtain a uniform drug concentration while sampling. Using a ±10% criterion as a means of determining drug degradation, drug concentrations were above 94% of initial values after 182 days in the refrigerator for all the samples. At room temperature, drug concentrations were above 98% of initial values at 119 days for both concentrations studied (Tables 3, 4; Figures 4, 5).

RESULTS (continued)

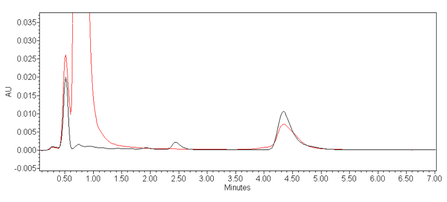

Figure 1. Sample chromatographic run of Clonidine Hydrochloride standard (black) and clonidine hydrochloride in PCCA SuspendIt (red) using an analysis wavelength of 220 nm.

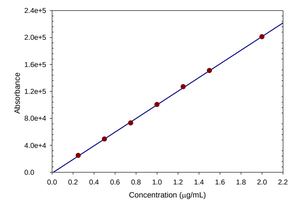

Figure 2. Calibration curve for high-performance liquid chromatographic analysis of Clonidine HCl (range: 0.25-µg/mL to 2.0-µg/mL). $r^2 = 0.9997$

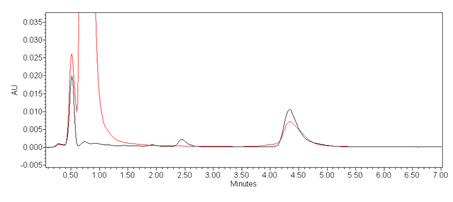

Figure 3. Chromatographic runs of Clonidine Hydrochloride in PCCA SuspendIt; standard (black), with caustic degradation (blue), acidic degradation (red), oxidative degradation (green), and ultraviolet light degradation (pink).

Table 1. Measurements of pH of Clonidine Hydrochloride in PCCA SuspendIt

Time	20 mcg/mL 5°C	20 mcg/mL 25°C	100 mcg/mL 5°C	100 mcg/mL 25°C
Day 0	5.14 ± 0.02	5.14 ± 0.02	5.12 ± 0.01	5.12 ± 0.01
Day 7	5.11 ± 0.01	5.10 ± 0.02	5.10 ± 0.01	5.09 ± 0.01
Day 14	5.12 ± 0.01	5.08 ± 0.04	5.09 ± 0.00	5.11 ± 0.01
Day 28	5.01 ± 0.02	5.03 ± 0.03	5.02 ± 0.02	4.95 ± 0.03
Day 42	5.09 ± 0.04	5.06 ± 0.02	5.06 ± 0.01	5.10 ± 0.05
Day 63	5.10 ± 0.03	5.09 ± 0.02	5.07 ± 0.01	5.05 ± 0.02
Day 91	5.07 ± 0.02	5.09 ± 0.03	5.08 ± 0.04	5.06 ± 0.02
Day 119	5.09 ± 0.02	5.10 ± 0.01	5.09 ± 0.01	5.08 ± 0.02
Day 182	5.14 ± 0.04	5.06 ± 0.14	4.74 ± 0.31	4.57 ± 0.09

Table 2. Viscosity (cP) measurements of Clonidine Hydrochloride in PCCA SuspendIt

Time	20 mcg/mL 5°C	20 mcg/mL 25°C	100 mcg/mL 5°C	100 mcg/mL 25°C
Day 0	44.6 ± 1.9	44.6 ± 1.9	45.3 ± 2.9	45.3 ± 2.9
Day 7	48.9 ± 5.8	46.1 ± 1.2	45.5 ± 1.4	45.2 ± 0.8
Day 14	49.9 ± 2.7	46.7 ± 2.1	47.6 ± 1.9	47.6 ± 1.6
Day 28	47.5 ± 2.2	44.6 ± 0.9	46.1 ± 0.4	44.1 ± 0.6
Day 42	47.2 ± 4.2	44.1 ± 0.9	44.9 ± 1.2	44.6 ± 2.6
Day 63	42.9 ± 1.3	44.2 ± 1.2	44.5 ± 1.1	42.8 ± 1.3
Day 91	48.4 ± 1.9	47.3 ± 1.9	47.9 ± 6.8	42.1 ± 5.2
Day 119	43.5 ± 2.7	44.1 ± 3.2	40.6 ± 3.2	43.2 ± 1.7
Day 182	41.8 2.4	43.5 ± 0.6	43.0 ± 0.4	48.5 ± 7.8

Table 3. Clonidine Hydrochloride Concentration (mcg/mL) in PCCA SuspendIt

Time	20 mcg/mL 5°C	20 mcg/mL 25°C	100 mcg/mL 5°C	100 mcg/mL 25°C
Day 0	19.81 ± 0.13	19.81 ± 0.13	100.38 ± 0.88	100.4 ± 0.88
Day 7	19.92 ± 0.20	20.23 ± 0.50	100.22 ± 0.16	99.64 ± 0.35
Day 14	19.74 ± 0.09	19.82 ± 0.34	100.25 ± 0.25	99.04 ± 0.85
Day 28	19.53 ± 0.27	19.46 ± 0.00	101.78 ± 1.90	99.79 ± 1.42
Day 42	20.00 ± 0.20	20.07 ± 0.06	99.34 ± 0.77	100.7 ± 0.27
Day 63	19.94 ± 0.13	19.51 ± 0.37	100.56 ± 1.05	100.2 ± 0.29
Day 91	19.27 ± 0.13	18.72 ± 0.14	99.61 ± 0.26	98.25 ± 0.47
Day 119	19.52 ± 0.26	19.83 ± 0.37	98.84 ± 0.89	98.37 ± 1.45
Day 182	18.74 ± 0.30	17.14 ± 0.42	99.08 ± 0.38	98.39 ± 0.59

Table 4. Percent of Clonidine Hydrochloride in PCCA SuspendIt Relative to Day-Zero concentration

Time	20 mcg/mL 5°C	20 mcg/mL 25°C	100 mcg/mL 5°C	100 mcg/mL 25°C
Day 0	100.0 ± 1.0	100.0 ± 1.0	100.0 ± 1.2	100.0 ± 1.2
Day 7	100.5 ± 1.2	102.1 ± 2.6	99.8 ± 0.9	99.3 ± 0.9
Day 14	99.6 ± 0.8	100.0 ± 1.9	99.9 ± 0.9	98.7 ± 1.2
Day 28	98.6 ± 1.5	98.2 ± 0.7	101.4 ± 2.1	99.4 ± 1.7
Day 42	100.9 ± 1.2	101.3 ± 0.7	99.0 ± 1.2	100.3 ± 0.9
Day 63	100.6 ± 0.9	98.5 ± 2.0	100.2 ± 1.4	99.9 ± 0.9
Day 91	97.3 ± 0.9	94.5 ± 0.9	99.2 ± 0.9	97.9 ± 1.0
Day 119	98.5 ± 1.5	100.1 ± 2.0	98.5 ± 1.2	98.0 ± 1.7
Day 182	94.6 ± 1.6	86.5 ± 2.2	98.7 ± 0.9	98.0 ± 1.0

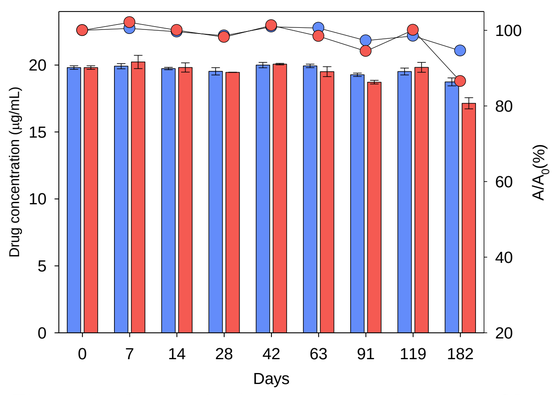

Figure 4. Change in drug concentration over 182 days on left axis (bars) for the 20-µg/mL samples of Clonidine Hydrochloride in PCCA SuspendIt stored at 5°C (blue) and 25°C (red); and relative change in percent on right axis (dots) as compared to initial concentration [A/Ao = drug content at time t (A) over initial drug content (Ao) x100].

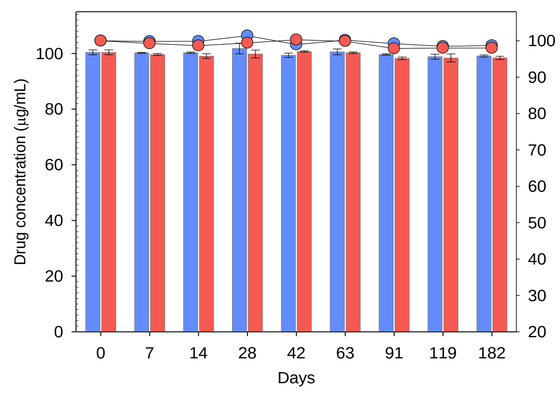

Figure 5. Change in drug concentration over 182 days on left axis (bars) for the 100-µg/mL samples of Clonidine Hydrochloride in PCCA SuspendIt stored at 5°C (blue) and 25°C (red); and relative change in percent on right axis (dots) as compared to initial concentration [A/Ao = drug content at time t (A) over initial drug content (Ao) x100].

CONCLUSIONS

A robust stability-indicating HPLC assay method for the determination of clonidine hydrochloride in PCCA SuspendIt was developed and validated. This assay was used to determine the chemical stability of the 20-µg/mL and 100-µg/mL concentrations of clonidine hydrochloride in PCCA SuspendIt at 5^0C and 25^0C. Drug concentration did not go below 98 % of the label claim (initial drug concentration) after 119 days at both concentrations and both temperature conditions studied. Given that the 20-µg/mL sample stored at room temperature did drop to 86.5%, a conservative BUD of 119 days across all concentrations and storage conditions is being recommended if refrigeration is not an option. The pH values did not change significantly, and while there was a slight decrease in the pH of the 100-µg/mL samples, it was not significant enough to affect the product stability. The viscosity of the suspensions was sufficient to allow easy re-dispersal of the drug particles upon shaking. Content uniformity was maintained, and no caking was observed. The preservative system in PCCA SuspendIt successfully protected the suspensions from growth of challenge microorganisms per the *USP* Chapter <51> AME Test. This study demonstrates that clonidine hydrochloride is physically, chemically, and microbiologically stable in PCCA SuspendIt for **182 days in the refrigerator** at both concentrations, and **119 days at room temperature** at both concentrations, thus providing a viable, compounded alternative for clonidine in a liquid dosage form, with an extended BUD to meet patient needs. The study further provides stability documentation over a bracketed clonidine hydrochloride concentration range between 20-µg/mL and 100-µg/mL, allowing compounding pharmacists more flexibility in customizing their formulations.

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